Preclinical Research Center
Ben-Gurion University of the Negev

STANDARD OPERATING PROCEDURE 102

FISH, AMPHIBIAN, AND REPTILE ANALGESIA

1. PURPOSE

This Standard Operating Procedure (SOP) intends to describe methods of assessing pain in fish, aquatic amphibians, and reptiles and mitigating pain by administering analgesic medications.

2. RESPONSIBILITY

Principal investigator (PI) and their research staff.

3. GENERAL CONSIDERATIONS

3.1. A procedure expected to be painful in humans is considered painful in animals.

3.2. When there is a question of whether a procedure is painful, the animal should benefit from analgesia.

3.3. Analgesia should be provided at an appropriate dose and frequency to control pain.

3.4. Any deviation from this procedure must be justified by the investigator and approved by the BGU ethical committee.

4. PAIN RECOGNITION AND ASSESSMENT

4.1. Adapt the frequency of observation to the invasiveness of the procedure (minimum once a day).

4.2. Start by observing the animal from a distance, so the observer's presence does not alter the animal's behavior. Then proceed to keep the animal more closely.

4.3. Look for any changes in the behavior. Report animals that appear to be in pain to the vets.

4.4. Although Fish, amphibians, and reptiles **do not** exhibit obvious clinical signs of pain, they can experience pain like mammals. Thus, the assumption is made by extrapolation from human observation.

Note: *The most reliable signs of pain and distress are changes in behavior.*

5. ANALGESIA PLAN

5.1. Specify the analgesia plan in your animal protocol.

5.2. Whenever possible, provide analgesia just before the painful stimulus, as it is more effective in preventing pain (e.g., give analgesic before surgery).

5.3. Use a combination of analgesics, often more effective than a single agent. For example, a variety of opioids, non-steroidal anti-inflammatory drugs (NSAID), and infiltration of a local analgesic.

6. LOCAL ANALGESIA

6.1. Infiltrate or apply local analgesics to areas where a painful stimulus is induced. Repeat the application of local agents at specified intervals to maintain analgesia. In some cases, a sedative is recommended when using regional analgesia.

Analgesic	Dose	Route	Duration	Note
Lidocaine	< 2 mg/kg	SC, Infiltration of surgical wounds	30–60 min.	Use lidocaine HCl 2% (20mg/ml) injectable solution. Because this drug is acidic, it is recommended to dilute it 3:1 with sodium bicarbonate injectable solution (at 5 or 8.4%). Dilution must be prepared immediately before use and should not be stored. A diluted solution is as effective, but induction of analgesia is slightly prolonged. *Dilution with sodium bicarbonate is unnecessary if lidocaine is administered to an anesthetized animal.
	EMLA cream	Topical	30–60 min.	Thick spread Apply only to intact skin. Shave or pluck the fur
	Localine			and apply a thick layer of cream, ideally 10 minutes before the painful procedure.
	Eye drops	Ocular	30-60min	
Bupivacaine	< 2 mg/kg	SC, Infiltration of surgical wounds	3–4 hrs.	Use bupivacaine HCl 0.50% (5mg/ml) injectable solution. Same comment as for lidocaine.

7. GENERAL ANALGESIA

7.1. Administration of non-steroidal anti-inflammatory drugs (NSAIDs):

7.1.1. NSAIDs include buprenorphine, carprofen, ketoprofen, and meloxicam.

7.1.2. To minimize chances for adverse drug interactions, a washout period of 5-7 days is recommended before switching between NSAIDs.

7.1.3. Ensure good water intake and monitor hydration status during the treatment period.

Fish

Analgesic	Dose	Route	Frequency	Note
Lidocaine	2-5 mg/L	Immersion	Immersion	
*MS222	25-300 mg/L	Immersion	Immersion	
Morphine	10 mg/ L	Immersion	Immersion	Controlled drug.

Amphibians

Analgesic	Dose	Route	Frequency	Note
Buprenorphine	75 mg/kg	SC	> 4 hr.	Controlled drug.

Reptiles

Analgesic	Dose	Route	Frequency	Note
*Buprenorphine	0.01 mg/kg	IM	q12/24 hr.	Controlled drug.
*Carprofen	2-4 mg/kg	PO, SC, IM	q24 hr.	Followed by 1-2 mg/kg q24-72 hr.
Ketoprofen	2 mg/kg	SC, IM	q24-48 hr.	
Meloxicam	0.1-0.2 mg/kg	PO, SC, IM	q24 hr.	

8. SAFETY PRACTICES

8.1. MS-222:

8.1.1. Wear protective clothing, gloves, and eye protection or work in a chemical fume hood when handling the MS-222 powder.

8.1.2. Wear gloves to handle animals exposed to MS-222